Serious Human Infections Due to Bacilli of the Arizona Group

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ORIGINALLY ISOLATED from sick Gila monsters, horned lizards, and a chuckawalla lizard in 1939, the first discovered strain of the Arizona group of enteric bacilli was considered to be an unusual salmonella variant.[3,10] Then, in the next few years, members of the Arizona group of organisms were identified in material from reptiles, birds, domestic animals and man throughout the United States and in every continent.[5,8,13] There is now abundant evidence that strains of these motile Gram-negative bacilli infect a variety of hosts, commonly invading the blood and producing diseases with high mortality rates in young animals and birds. Particularly troublesome economically have been multiple outbreaks of bird septicemia and diarrhea in turkey farms in California and other states. Loss of as much as 60 per cent of a flock of young turkeys has been reported. These outbreaks have been carefully traced to commercial hatching eggs and to dried egg powder.[7]

Largely through the work of Edwards and his co-workers the Arizona group has been separated from the multitude of enteric bacilli and has been found to comprise a compact biochemical group of bacteria related to both coliforms and salmonellas.[4,6,7,13] They differ enough serologically and biochemically, however, to be regarded by authorities in this field as a separate individual genus, of which approximately 150 serotypes are now recognized.[5,11] Now that these organisms have been more clearly defined, it is no longer appropriate to place them in the vague category of "paracolons."

Infections in man are manifested by a variety of clinical forms, in general resembling salmonella infections. The cases recorded have been even somewhat more severe than the usual cases of salmonellosis, although more experience is needed to determine whether this apparent severity reflects factors of selectivity in recognizing Arizona infections. Mass infections from ingestion of food, with vomiting, diarrhea and fever, have occurred, in which Arizona strains were incriminated. Some of these outbreaks were traced to food such as ice cream and cream pie and to food handlers.[6,12] Typhoidal syndrome has been observed.

Certain serological strains appear to have a propensity for producing localized infections or abscesses. Strains of the commonest serotype in human infections (7:1, 2, 6) appear to be especially invasive. Over half of the strains of this serotype were isolated from infections of organs and tissues other than the intestinal tract in the series of Edwards, McWhorter and Fife.[6] The strains isolated in both of the cases reported herein are members of this serotype (7:1, 2, 6). This type is widely distributed in fowls in California.

Various localized human infections from which members of the Arizona group have been isolated include brain abscess, pleuritis, scalp abscess, conjunctivitis, osteomyelitis,[9] otitis media[2] and liver abscess.

Five deaths apparently resulting from infection by Arizona strains have been listed by Edwards, McWhorter and Fife[6]: (1) A 58-year-old man with severe gastroenteritis. (2) An undescribed patient with an hepatic abscess. (3) A 5-month-old infant with diarrhea. (4) A 5-month-old infant from which a member of the Arizona group was cultured from cardiac blood. (5) One of the cases (Case 1) reported herein.

In spite of the accumulating references to cases of serious human infection in public health reports and microbiological journals, few detailed clinical studies[8,9,12,14] and no detailed autopsy reports have been published.

In the two following cases the chief pathologic lesions consist respectively of (1) a fatal hepatic abscess, and (2) chronic osteomyelitis of the left femur with septic arthritis of the knee joint. In both cases bacilli of the Arizona group were isolated in pure culture from the lesions.

REPORT OF CASES

CASE 1. An 87-year-old white man, was admitted to Santa Clara County Hospital from a lodge rest home where he had been receiving domiciliary care. On the day before admission, convulsions developed unexpectedly. When a physician who was called noted that the temperature was 104°F. and that glycosuria was present, the patient was sent to Santa Clara County Hospital. Upon examination there, the patient was observed to be obese, senile, confused, lethargic and febrile. Abdominal guarding was noted. Neither jaundice nor diarrhea was present.

Leukocytes numbered 11,400 per cu. mm., of which 90 per cent were neutrophils. The blood sedimentation rate was 31 mm. in one hour (Westergren). Blood glucose was 330 mg. per 100 cc. Urinalysis showed 4 plus glycosuria and 2 plus acetonuria. A culture of blood grew no organisms. An area of patchy density in the base of the left lung was observed in an x-ray film of the chest.

The hyperglycemia and mild acetonuria were easily controlled by intravenous fluids and relatively small doses of regular insulin. However, the patient remained stuporous and confused. A spinal puncture was done the day after admission and the initial pressure was 200 mm. of water. The spinal fluid was clear and colorless and contained 4 leukocytes per cu. mm. Cultures of spinal fluid grew no organisms. On the day following admission, the serum sodium was 130 mEq. per liter, the potassium was 5.1 mEq./l. and the CO_2 was 29 mEq./l. The tem-

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perature remained elevated, going as high as 104.4°F. (rectal). Hiccupping was an almost continual problem. The patient grew weaker without improvement and died four days after admission. Therapy from the time of admission consisted of penicillin (600,000 units intramuscularly daily), streptomycin (0.5 gm. initially and 0.5 gm. every 6 hours intramuscularly), digoxin (0.5 mg. daily), continual administration of oxygen nasally, and fluids intravenously and subcutaneously.

At autopsy five hours after death, on gross examination three apparently healing, superficial ulcers were noted on the left posterior trunk and there was a similar ulcer over each lateral malleolus. The liver weighed 2030 gm. and was reddish-brown, smooth and glistening. On the superior surface over the right lobe adjacent to the diaphragm was an irregular yellowish area of softening, 10 cm. across. Beneath this area, on sectioning, a large irregular abscess up to 12 cm. was exposed (Figure 1). The abscess was poorly demarcated, multiloculated and filled with yellow-gray viscid purulent material, with several indefinite smaller necrotic foci surrounding the abscess in a patchy distribution. The remainder of the liver substance was uniformly reddish-brown. The gallbladder was scarred and contracted about a single large 3 cm. calculus. No evidence of acute inflammatory reaction was noted in the gallbladder or biliary ducts. The lungs were hyperemic and showed evidence of patchy bronchopneumonia. The heart was moderately concentrically enlarged to 500 gm. The myocardium showed no changes, and there was moderate (Grade II-III) atherosclerosis of coronary arteries. The valves showed no important changes. The spleen was enlarged to 320 gm. and it was soft and mushy, the cut surfaces bulging. The pancreas appeared normal. No significant changes were observed in the entire gastrointestinal tract. Except for a moderate degree of cortical scarring no abnormality was seen in the kidneys. Upon microscopic examination of the liver a multiloculated abscess composed of necrotic hepatic cells and polymorphonuclear exudate was observed. Patchy collections of polymorphonuclear cells were scattered throughout the adjacent hepatic parenchyma (Figure 2). Gram-negative bacilli were easily seen in the abscess. Culture of material from the abscess grew bacilli of the Arizona group. Foci of acute bronchopneumonic exudate were present in the lungs. The splenic red pulp was hypercellular and contained numerous polymorphonuclear and plasma cells. The gallbladder wall was thickened and scarred, without evidence of acute inflammation. Sections of both kidneys showed moderate arterial and arteriolar hypertensive changes. Distinct microscopic stigmata of diabetes mellitus as described by Bell[1] were not found in the kidney or pancreas.

No clinical or autopsy evidence was found to suggest the manner of development of the liver abscess. It seems unlikely that the contracted gallbladder with its calculus was a primary portal of entry. The small skin ulcers also appear unlikely.

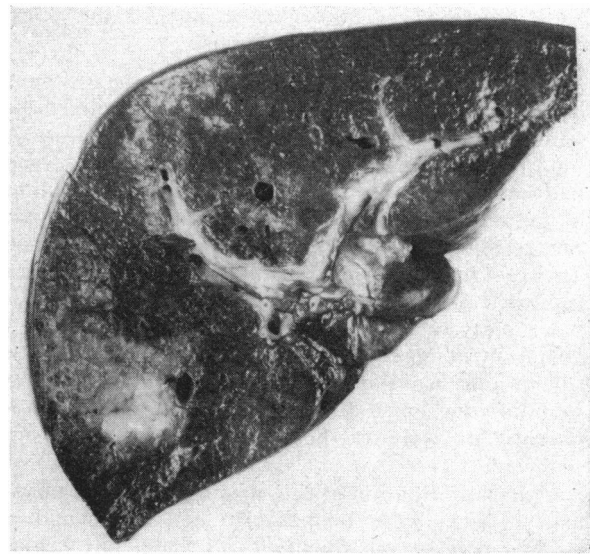

Figure 1 (Case 1).—Gross view of liver, showing irregular multiloculated abscess in right lobe containing yellow-gray viscid purulent material. Several indefinite smaller necrotic foci surround the abscess in a patchy distribution.

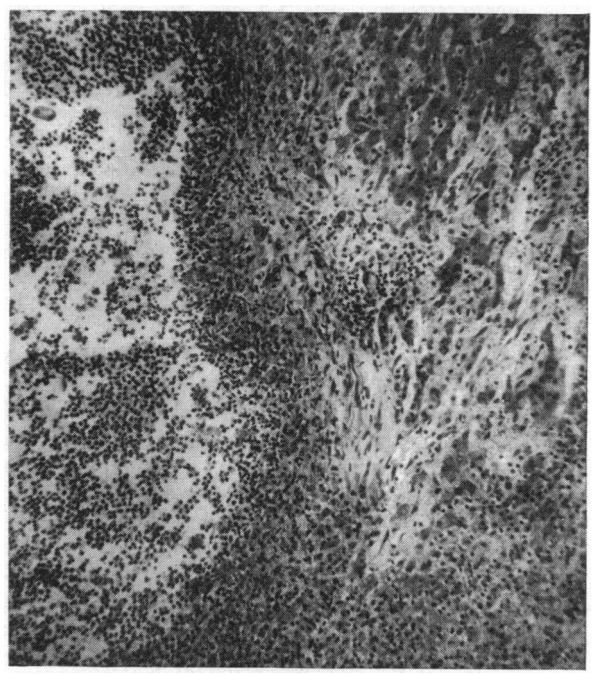

Figure 2 (Case 1).—Microscopic section of liver showing abscess containing acute polymorphonuclear exudate and adjacent necrotic hepatic parenchyma. (Magnification ×80).

No bacteriologic investigation was made of the environment from which the patient was admitted, but it appears most likely that the infection was acquired from contaminated food. The significance of hyperglycemia and glycosuria in this case is conjectural. It may be stated that pyogenic abscesses are not a common complication of diabetes mellitus. From the other point of view, diabetes mellitus has not

been described as occurring in any of the other case reports of Arizona infections.

CASE 2. A 63-year-old white housewife was sent by a physician to Santa Clara County Hospital September 2, 1958, because of fever and painful swelling of the left knee. For the previous ten months she had noted gradually increasing aching and swelling of the left knee, which was aggravated by walking. She recalled no significant trauma to the knee. One week before admission the referring physician had aspirated some fluid from the knee joint. The swelling rapidly recurred. Serious past illnesses included acute thrombocytopenic purpura in 1947, for which splenectomy was done, apparently with cure. In 1950 jaundice developed for a short time. This was apparently due to viral hepatitis, and there were no sequelae.

Upon examination at this hospital the chief physical findings were temperature of 103°F. and a swollen, tender and fluctuant left knee. The hemoglobin was 9.2 gm. per 100 cc. and the hematocrit 32 per cent. Leukocytes numbered 16,950 per cu. mm., of which 78 per cent were neutrophils. The urine was normal except for a trace of albumin. The serum albumin was 3.3 gm. per 100 cc., the globulin 3.1 gm. per 100 cc. and the serum total bilirubin 0.9 mg. per 100 cc. The serum VDRL was reactive. Several abnormalities were observed in the left femur in x-ray films taken at the time of admission. These consisted of an irregular sclerosis at the margins of the lower third of the shaft of the femur with periosteal proliferation. Several oval radiolucent areas within the shaft of the femur suggested localized bone absorption. There was no break in the cortex. The appearance was that of chronic osteomyelitis involving the lower third of the femur and its periosteum.

On the day the patient was admitted the knee was aspirated and 450 cc. of thick fluid resembling tomato soup was withdrawn. Cultures of this fluid grew bacilli of the Arizona group. On the sixth day after admission the focus of osteomyelitis was curetted through a 2.5 x 1.5 cm. window in the cortex of the lateral aspect of the distal femur. The cortex had a rough chalky appearance. A copious amount of whitish-yellow cheesy material was curetted from the marrow cavity. No pus was noted in the marrow cavity. Microscopic sections of tissue taken from the knee joint showed nonspecific chronic inflammatory granulation tissue of no distinctive pattern.

Chemotherapy consisted of achromycin orally, 250 mg. four times a day for a prolonged period. The patient had a spiking fever at first, the temperature going up to 103°F. The temperature gradually subsided in the ensuing three weeks and then remained within a normal range. The surgical wound continued to drain fluid, however, and no improvement of the bony lesions was seen radiographically. A more extensive curettage of the medullary canal was performed December 3, 1958. Multiple cultures of curetted material grew Arizona bacilli of the strain previously isolated from the knee joint effusion.*

BACTERIOLOGIC OBSERVATIONS

Bacilli of the Arizona group were isolated in pure culture from infected material in both cases. In Case 1 direct smears of pus from the liver abscess taken at autopsy showed Gram-negative bacilli similar to coliforms. Pus was inoculated on eosin-methylene-blue agar, Salmonella-Shigella agar and nutrient blood agar plates, and small colorless colonies grew overnight on all plates. Cultures of these were subsequently identified as members of the Arizona group by Dr. P. R. Edwards at the Communicable Disease Center at Chamblee, Georgia. In Case 2 bacilli of the Arizona group were repeatedly isolated in pure culture from fluid aspirated from the left knee joint and from infected necrotic material curetted from the medullary canal of the left femur. Cultures of these organisms were also identified by Edwards. In both cases the strains were found to be members of the same serologic type (7:1, 2, 6).

Subcultures of both strains had the following fermentation reactions: Lactose, no fermentation at 24 hours. (Acid was produced by subcultures of the strain of Case 1 after several transfers); salicin, no fermentation; sucrose, no fermentation; maltose, acid and gas; mannitol, acid and gas; dextrose, acid and gas; triple sugar iron agar, alkaline slant with acid, gas and H_2S in the butt. The organisms were motile, were negative for indole production and had no hydrolytic effect on urea. They caused a slight liquefaction of gelatin after ten days. There was positive reaction to methyl red but not to a Voges-Proskauer test.

Following the lead of Murphy and Morris,[12] who found that their infected patients developed agglutinating antibodies against the infecting Arizona strain, we tested the serum of the patient of Case 2 for the presence of antibodies against both Arizona strains which had been isolated in Case 1 and Case 2. Her serum was found to agglutinate Arizona bacilli isolated from her leg lesion to a serum dilution of 1:1280. In addition, serum from that patient agglutinated bacilli of the strain which had been isolated from the liver abscess in Case 1 to a serum dilution of 1:640. As a check against nonspecific cross reaction, serum of the patient in Case 2 was tested for antibodies against S. typhosa (H and O), S. paratyphi A, S. paratyphi B, and P. tularensis, and no agglutination was demonstrated against these.

DISCUSSION

The emergence of a newly recognized group of pathogenic enteric bacilli introduces several problems. It is apparent that bacterial pathogenicity is a relative quality, ranging at one extreme from the ability to develop progressive invasion of the body

*The patient died on February 6, 1959, 30 hours after another extensive sequestrectomy of the left femur. At autopsy there was bilateral bronchopneumonia. Organisms of the Arizona group were isolated at autopsy from the medullary cavity of the left femur but not from the lungs or blood.

when introduced by any route and at the other extreme to only a weak ability to multiply and grow when introduced directly into tissues. The ability of a given strain of enteric bacilli to produce symptoms in most of the individuals infected varies decidedly in both the species of hosts infected and the individuals infected. The evidence that members of the Arizona group are primary incitants of disease when ingested is very strong. The epidemiology of the infections in fowls has been particularly clear-cut, and there have been repeated instances in which it was possible to trace the transmission of infection through eggs.[5,7] Multiple outbreaks of severe gastroenteritis in man in which epidemiologically significant Arizona strains were isolated from the affected persons and not from asymptomatic persons have been well documented. Following the infections, the patients developed antibodies against the corresponding Arizona strain.[12] In some of these outbreaks the corresponding Arizona strain was isolated from food or food handlers. As a corollary to this, Arizona strains are rarely isolated from asymptomatic persons. Direct evidence of their ability to invade and multiply is supplied by the isolation of organisms from primary abscesses or other localized infections unassociated with the presence of foreign material in tissue and unassociated with other known pathogenic organisms.

Clinicians may well wonder why a group of organisms related to salmonellas and similar in their pathogenicity is not simply classified as a salmonella. The viewpoint of experienced workers in the isolation and epidemiology of enteric organisms is stated by Edwards, Cherry and Bruner[4]: "Without clear concise generic definitions [of salmonellas] the worker in a diagnostic laboratory is at a decided disadvantage. To relax the generic definition would lead inevitably to placing many atypical coliforms of doubtful pathogenic significance in that genus." For these reasons the Arizona species were placed in a separate genus.

Although they usually grow well on ordinary laboratory media used to isolate enteric pathogens, members of the Arizona group are difficult to recognize in the clinical laboratory and can be identified only by complex serological typing in special salmonella typing centers. In the clinical laboratory they are most likely to be confused with salmonellas, an error which is not especially serious. A much more important problem of recognition is encountered when an unrecognized Arizona strain ferments lactose early—and many strains apparently have this property.[15] The reason for this is that it is almost universal practice in clinical laboratories to consider a culture of an enteric bacillus which ferments lactose to be a nonpathogenic organism and to report it to the clinician as a coliform or paracolon. This reliance on lactose fermentation to screen out potentially dangerous organisms is, of course, very useful in general diagnostic bacteriological practice, but permits an unknown number of lactose-fermenting Arizona strains, as well as epidemiologically significant strains of E. coli, to go unrecognized.

An improved method for early recognition of this group in the clinical laboratory is needed.

SUMMARY

Bacilli of the Arizona group were isolated in pure culture of material from a fatal case of acute liver abscess in an 87-year-old white man and from a 63-year-old white woman who had chronic osteomyelitis of the left femur with septic arthritis of the knee joint.

The Arizona genus comprises a group of widely distributed pathogenic Gram-negative enteric bacilli which are related to both salmonellas and coliforms. Arizona strains have produced serious human infections with a high degree of invasiveness and occasional death. Members of this group are easily isolated and grown in the clinical laboratory, but are difficult to recognize.

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